

Fig. 1

5'  
 GTG GAA TTC TGC AGA TGC TAC CGG ACT CAG ATC AAT TCA CAT CCA CCA GCC  
 ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG  
 M R V L V L A L A V A L A V G D Q  
OaVtgss Cleavage site  
 TCC AAC TTG GGG GAT CTA GGC TTG TGT GAT GAA ACG AGG TTC GAG TGT AAG  
 S N L G D L G L C D E T R F E C K  
 Factor C  
 TGT GGC GAT CCA GGC TAT GTG TTC AAC ATT CCA GTG AAA CAA TGT ACA TAC 3'  
 C G D P G Y V F N I P V K Q C Y F

**pAc5/VtgCrFCES-V5-His**

Fig. 2A

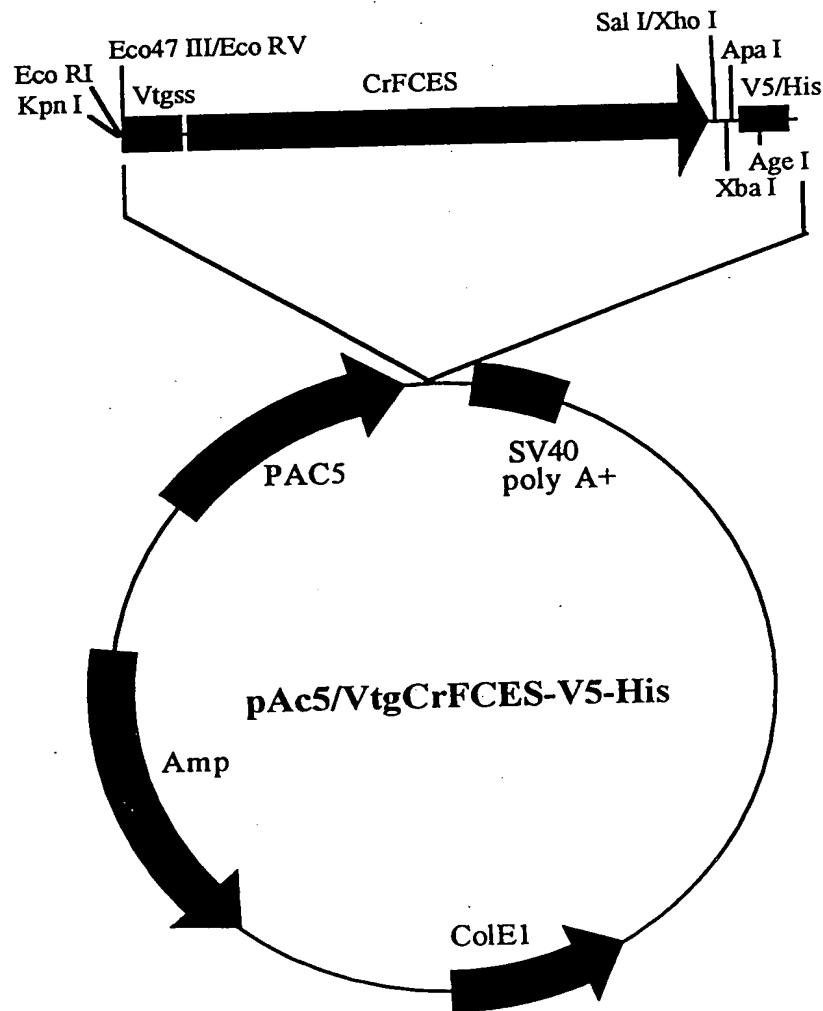
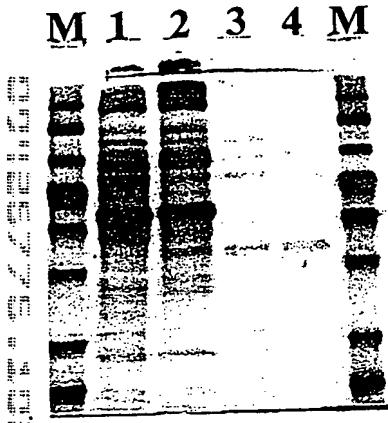


Fig. 2B

Coomassie Blue-stained  
SDS-PAGE



Western Blot using  
INDIA-His-HRP

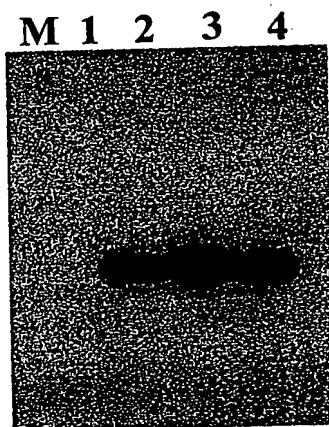


Fig. 3A

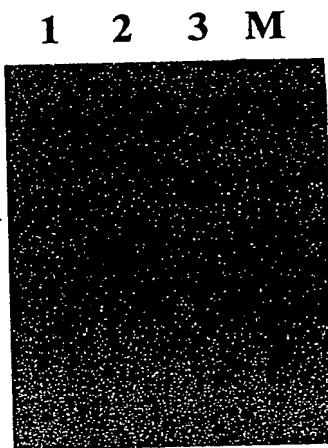
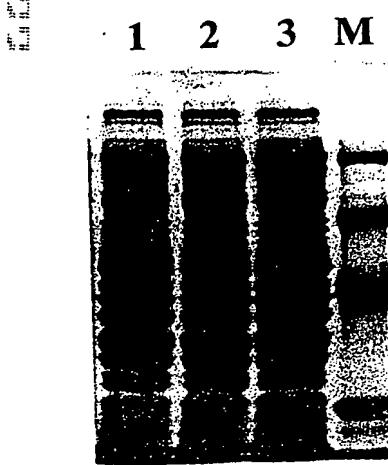


Fig. 3B

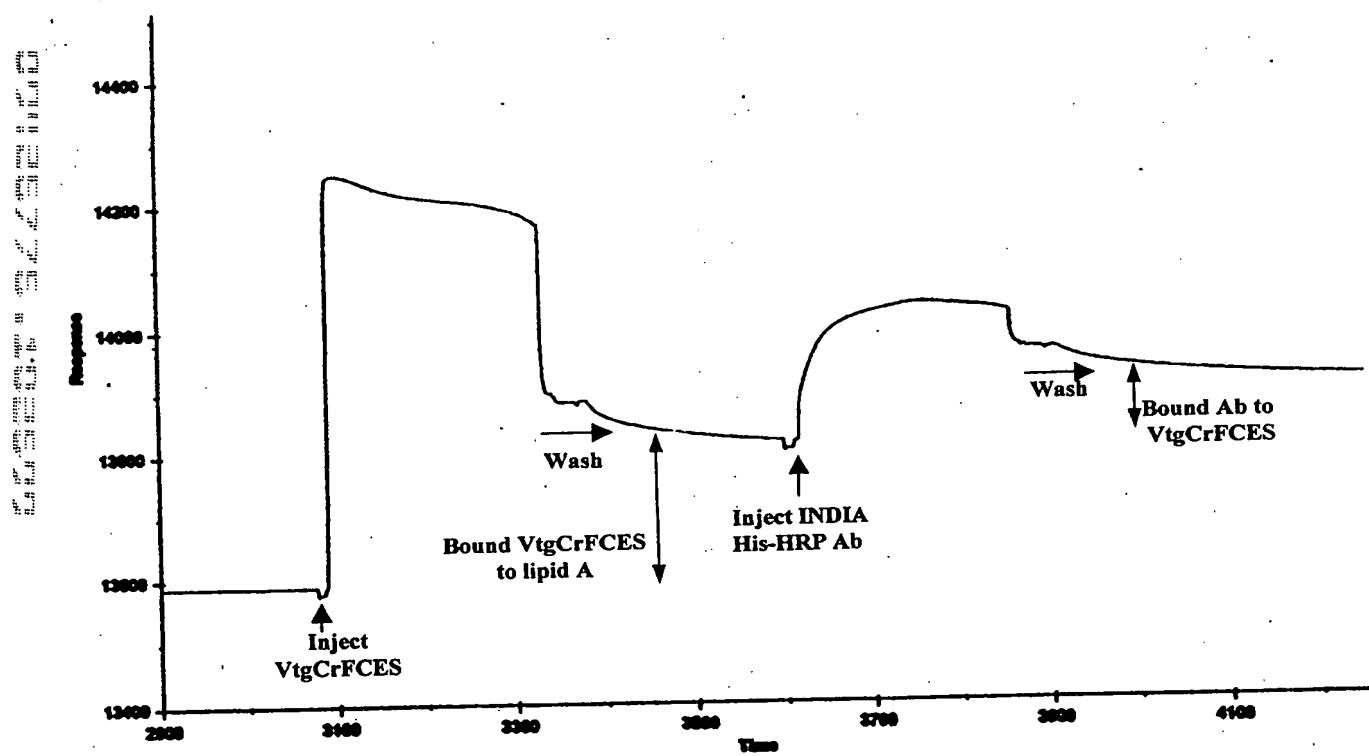


Fig. 4

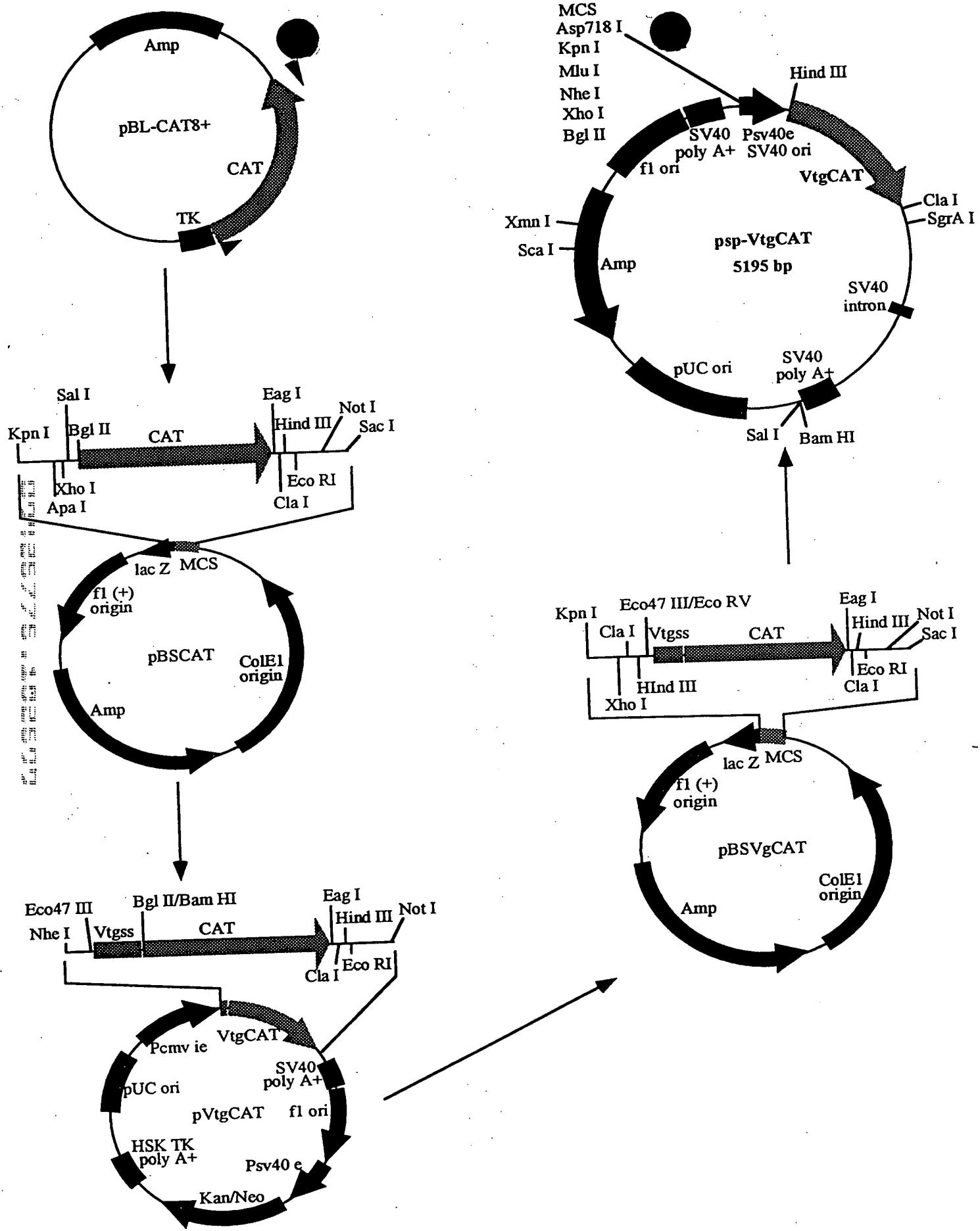


Fig. 5A

5'

Hind III Eco RV/Eco 47III  
 ATC GAT AAG CTT GAT GCT ACC GGA CTC AGA TCA ATT CAC ATC CAC CAG CC  
 ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG  
 M R V L V L A L A V A L A V G D Q  
 → OaVtgss

Bgl II/Bam HI  
 TCC AAC TTG GGG GAT CTG CTG GAG AAA AAA ATC ACT GGA TAT ACC ACC GTT  
 S N L G D L L Q K K V T G W T T V  
 → CAT

Eag I  
 ... ... ... GGC GGG GCG TAA TTT TTT TAA GGC ACG GCC GAT GCG ACG  
 G G A \*\*\*

Cla I  
GTA TCG ATA ACT TGA TAT CG 3'  
 Hind III

Cleavage site

pBSVtgCAT

Fig. 5B

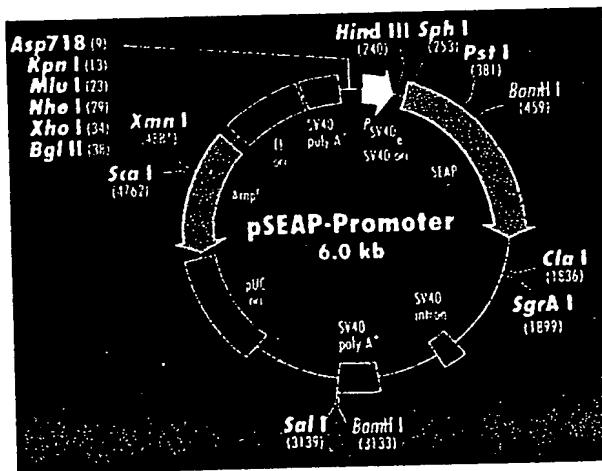


Fig. 5C

5'

Hind III Eco RV/Eco 47III  
 TGC AAA AAG CTT GAT GCT ACC GGA CTC AGA TCA ATT CAC ATC CAC CAG CC

ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG  
 M R V L V L A L A V A L A V G D Q

OaVtgss → Cleavage site

Bgl II/Bam HI  
 TCC AAC TTG GGG GAT CTG CTG GAG AAA AAA ATC ACT GGA TAT ACC ACC GTT  
 S N L G D L L Q K K V T G W T T V

CAT →

Eag I  
 ... ... ... GGC GGG GCG TAA TTT TAA GGC ACG GCC GAT GCG ACG  
 G G A \*\*\*

Cla I  
 GTA TCG ATA TTG TTA CAA CAC CCC AAC 3'

ssp-VtgCAT

Fig. 5D

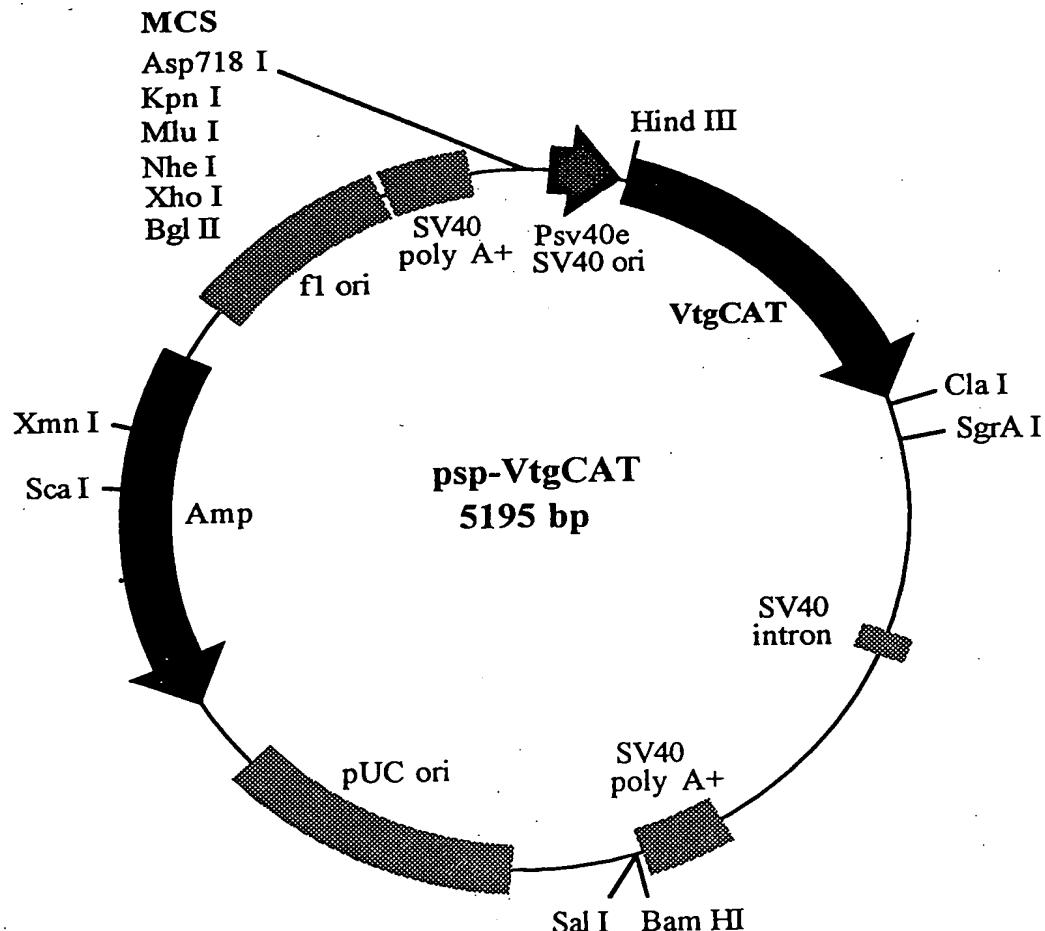


Fig. 5E

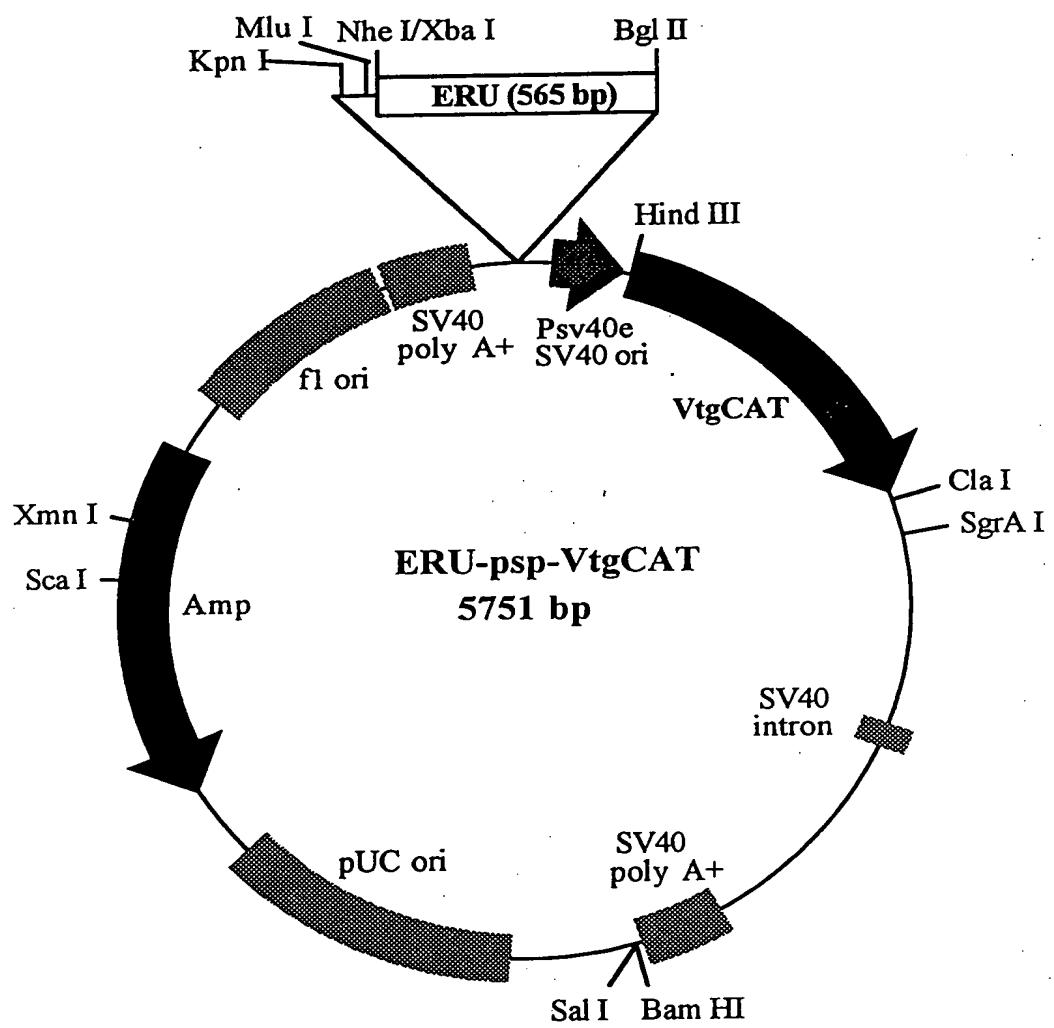


Fig. 5F

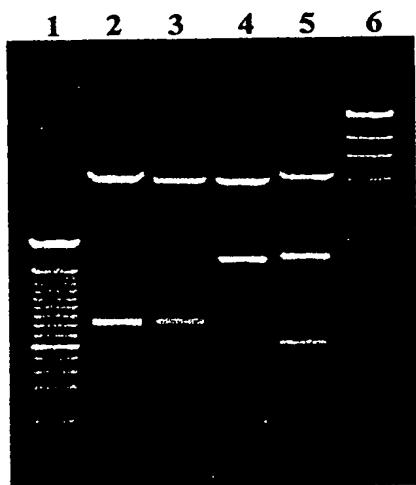
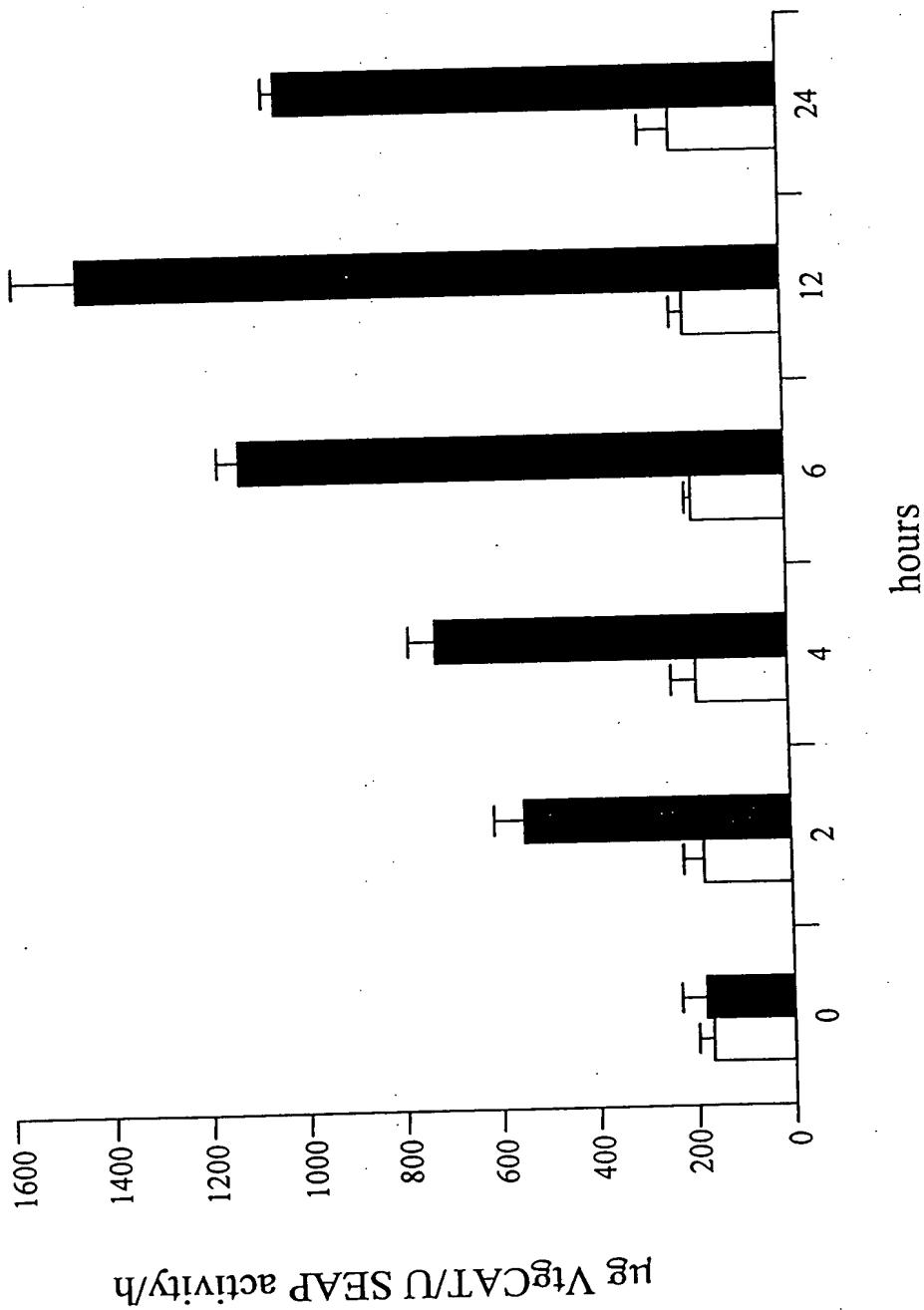


Fig. 56

Fig. 6



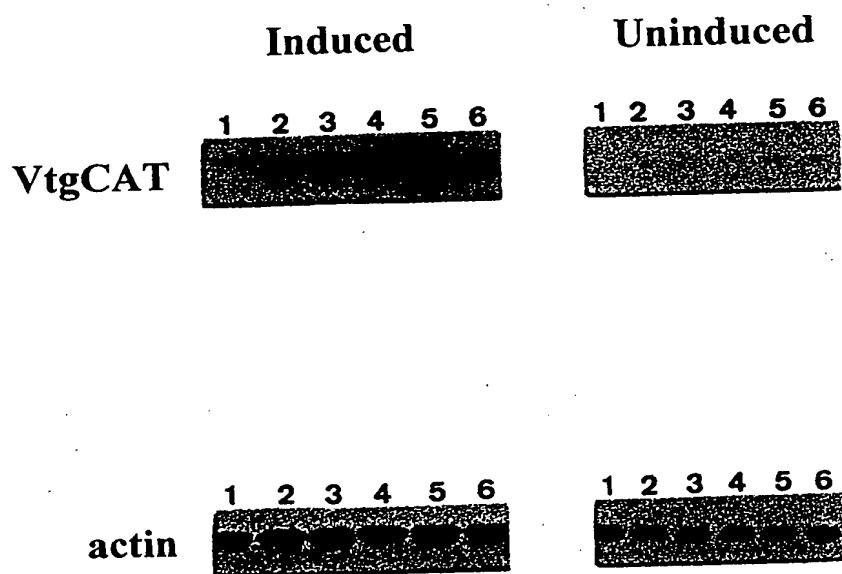


Fig. 7

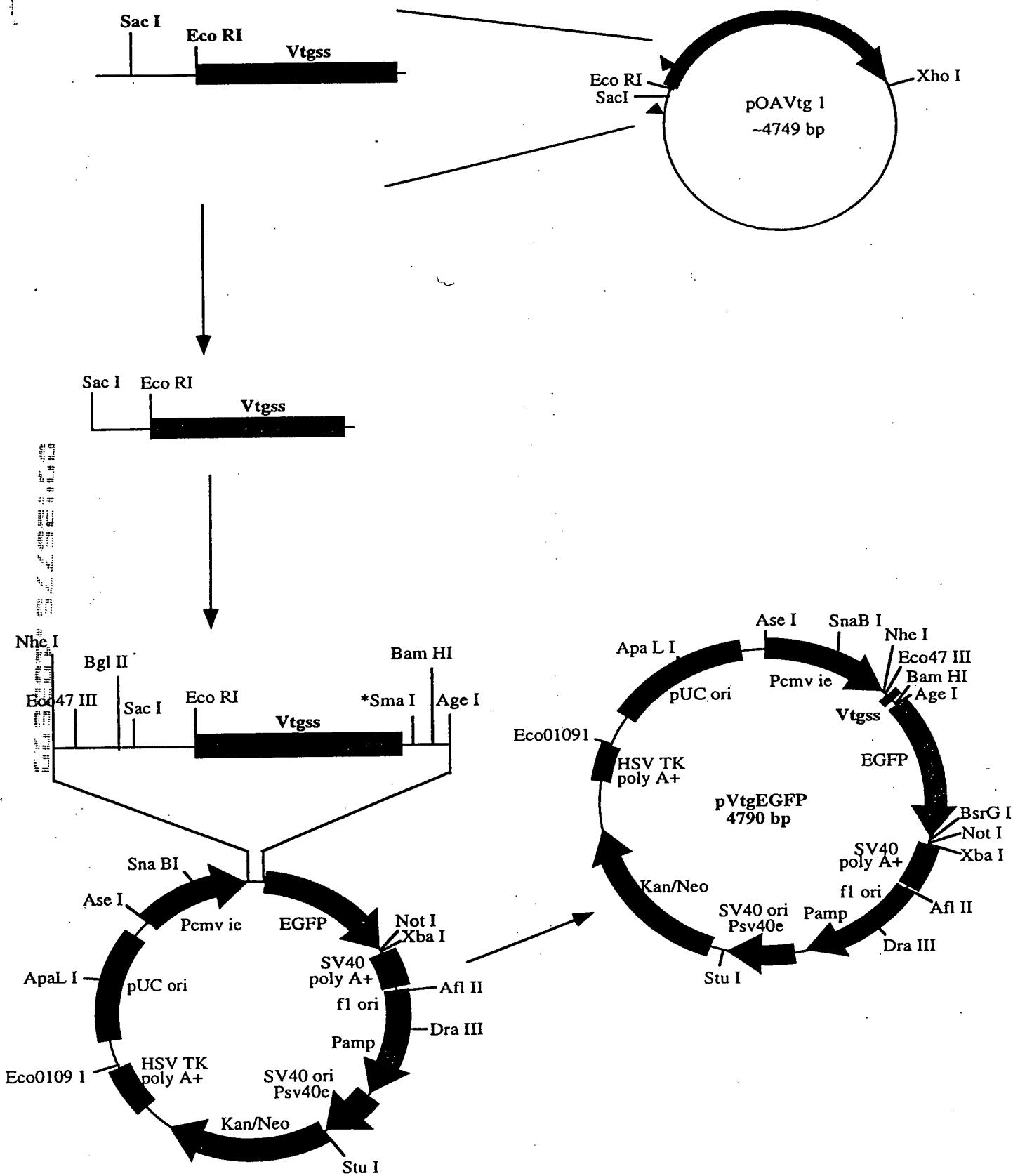


Fig. 8A

5'

Nhe I +1  
 GCT AGC GCT ACC GGA CTC AGA TCA ATT CAC ATC CAC CAG CC  
 Eco 47 III

ATG AGG GTG CTT GTA CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG  
 M R V L V L A L A V A L A V G D Q  
OaVtgss →

Bam HI Age I  
 TCC AAC TTG GGG GAT CCA CCG GTC GCC ACC ATG GTG AGC AAG GGC GTG GTG  
 S N L G D P P V A T M V S K G V V  
EGFP →

CAG AAC TCC GGG 3'  
 Q N S G

**pVtgEGFP**

Fig. 8B

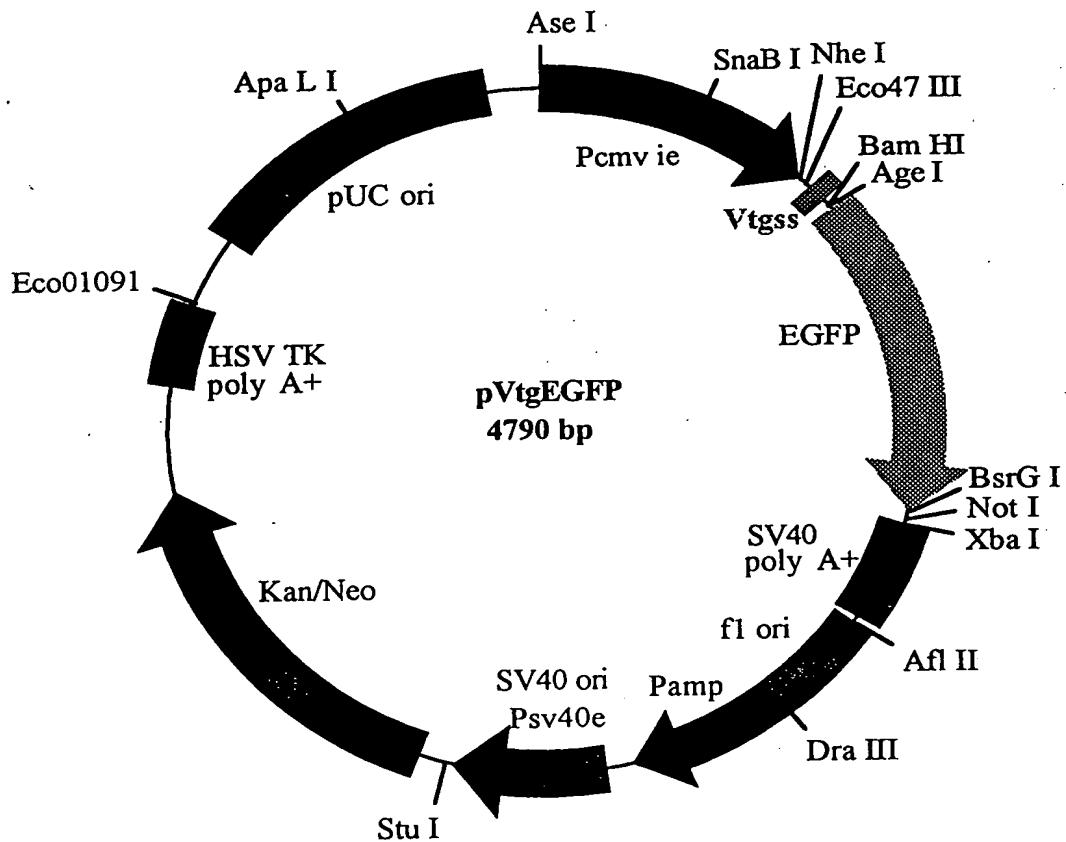


Fig. 8C

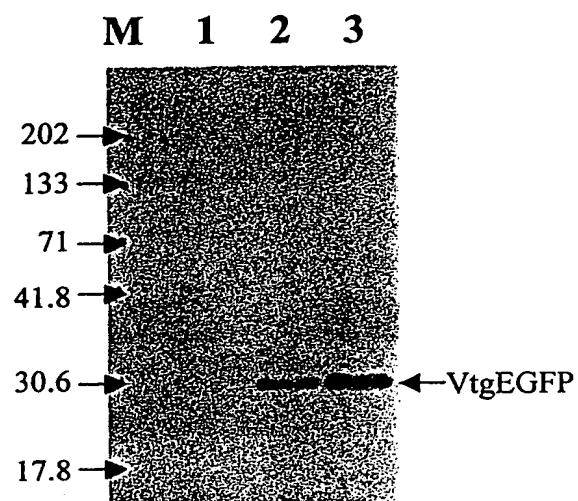
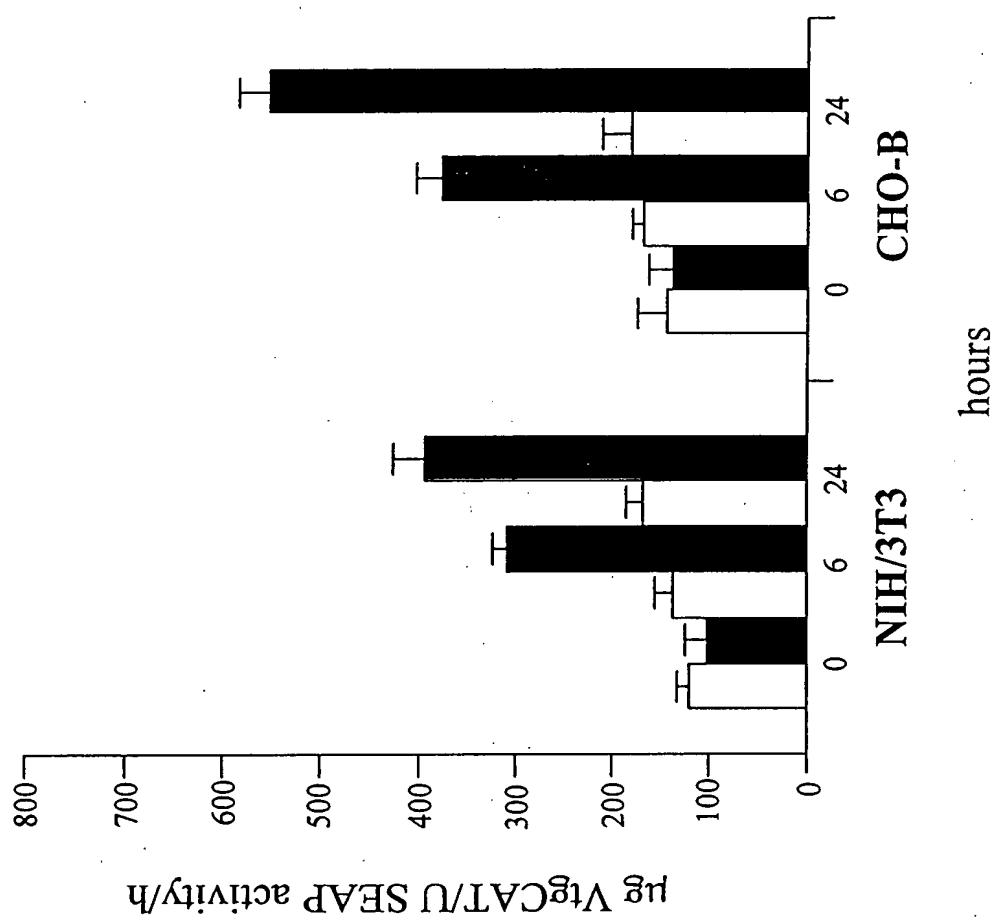


Fig. 8D

Fig. 9



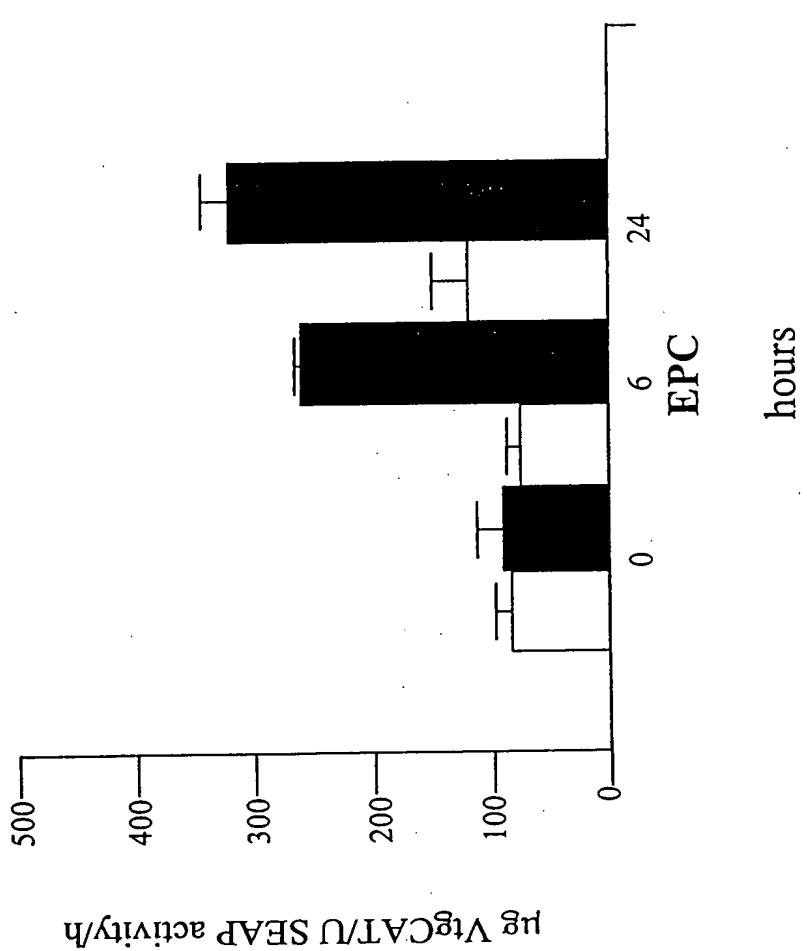


Fig. 10

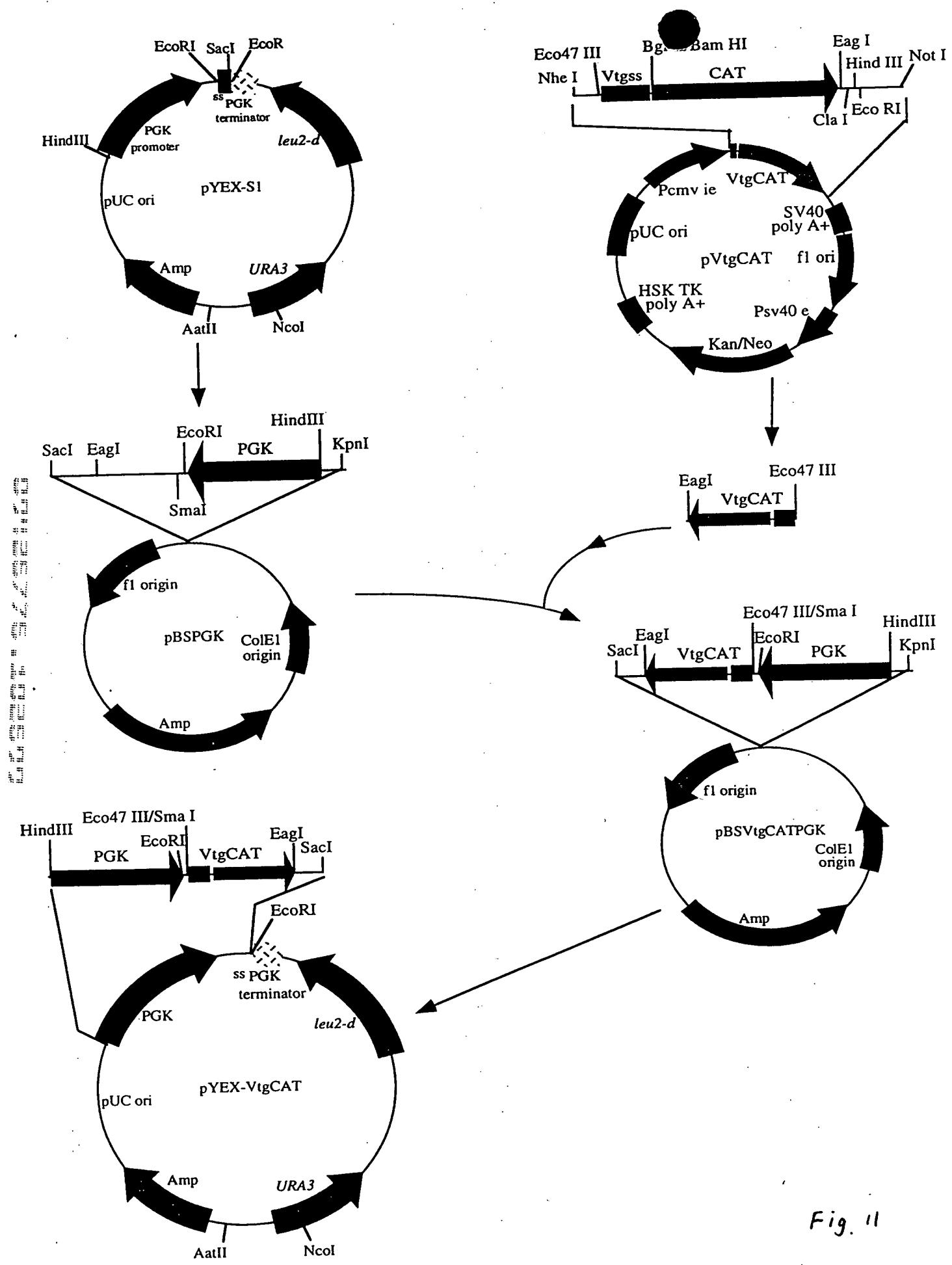


Fig. 11

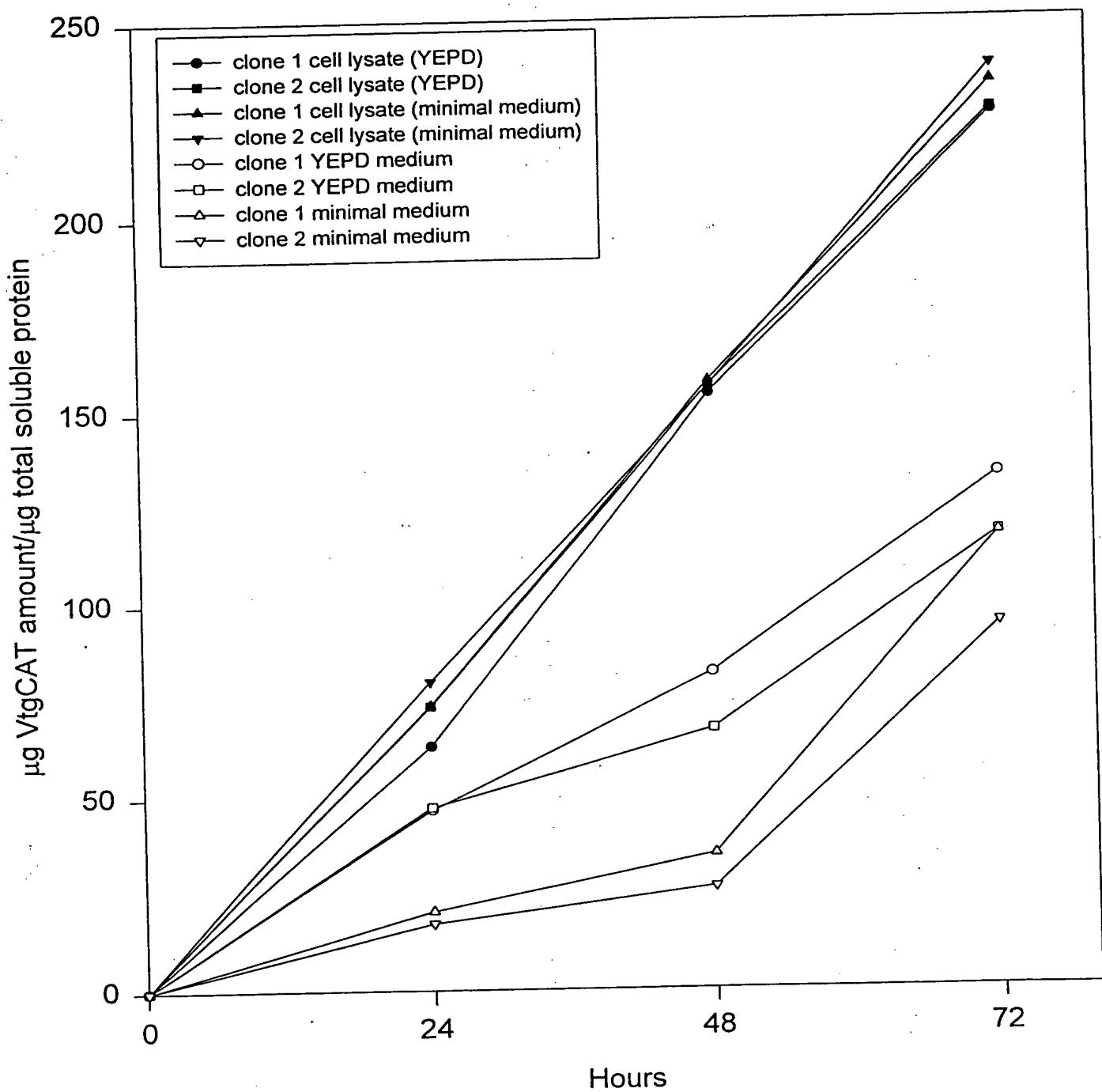


Fig. 12

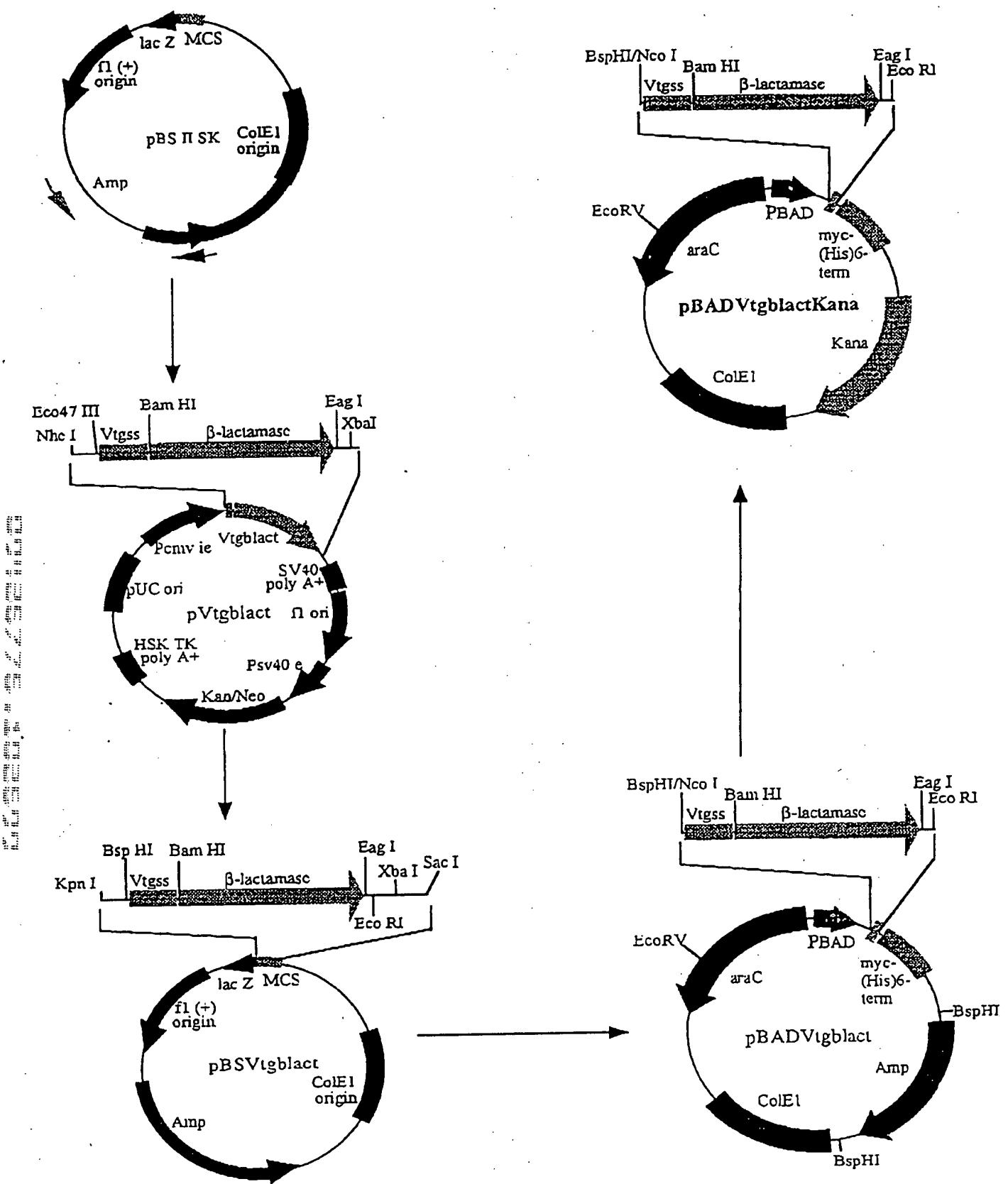


Fig. 13

5'  
 CTC TAC TGT TTC TCC ATA CCC GTT TTT TTG GGC TAA CAG GAG GAA TTA ACC  
 ATG AGG GTG CTT GTC CTA GCT CTT GCT GTG GCT CTC GCA GTG GGG GAC CAG  
 M R V L V L A L A V A L A V G D Q  
 OaVtgss → Cleavage site  
 Bam HI  
 TCC AAC TTG GGG GAT CCA GAA ACG CTG GTG AAA GTA AAA GAT GCT GAA GAT  
 S N L G D P E T L V K V K D A E D  
 β-lactamase  
 CAG TTG GGT GCA CGA GTG GGT TAC ATC GAA CTG GAT CTC AAC AGC GGT AAG 3'  
 Q L G A R V G Y I E L D L N S G K

**pBADVtgblactKana**

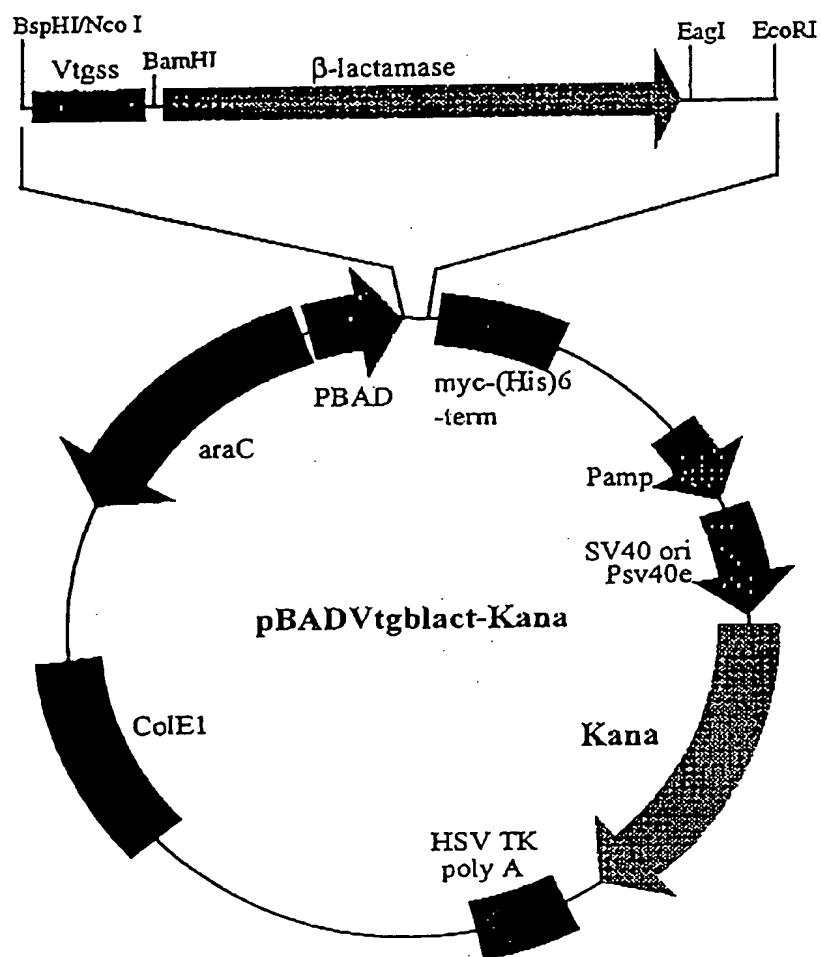


Fig. 14 B

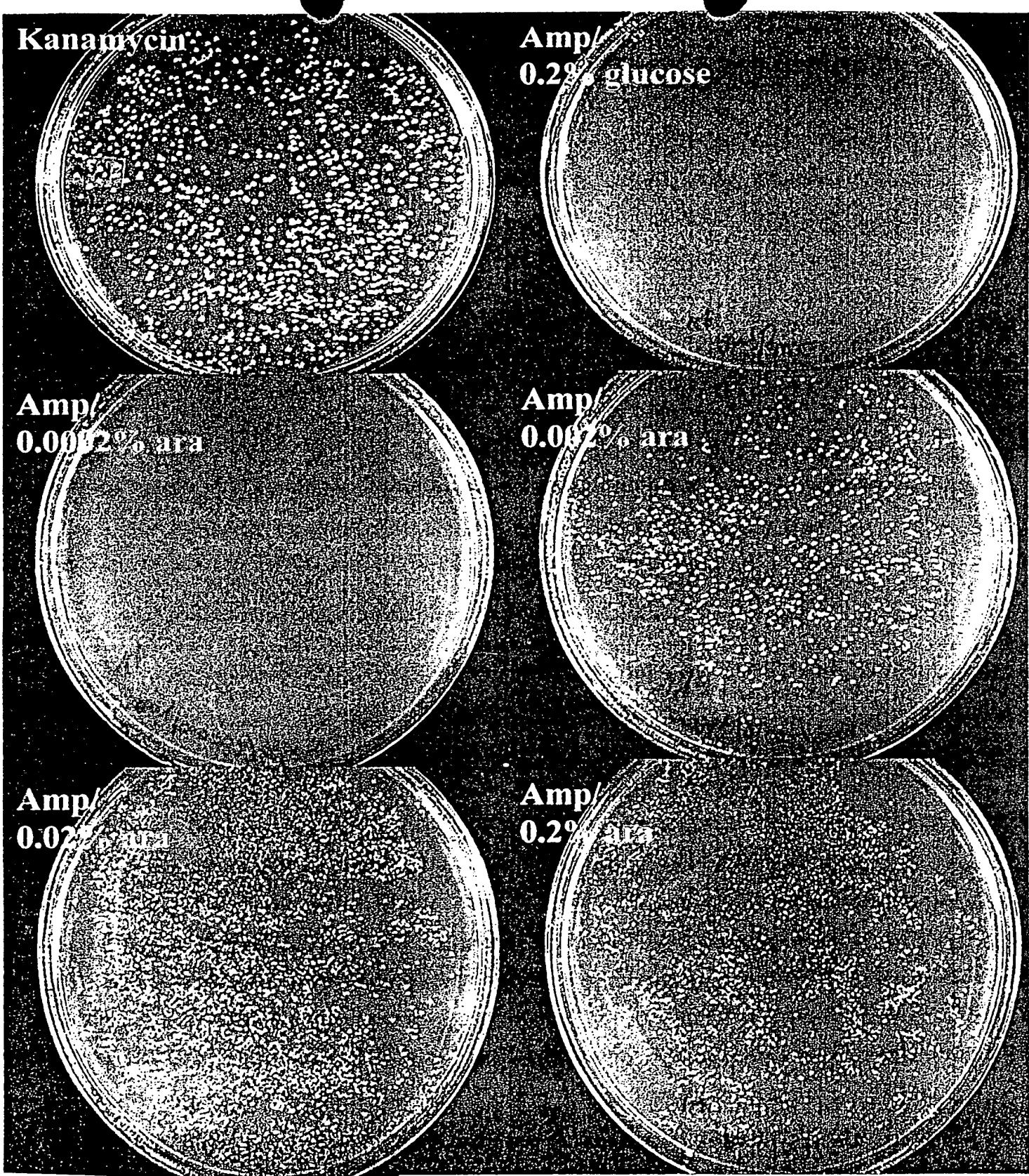


Fig. 15

### Growth profile of bacteria after induction

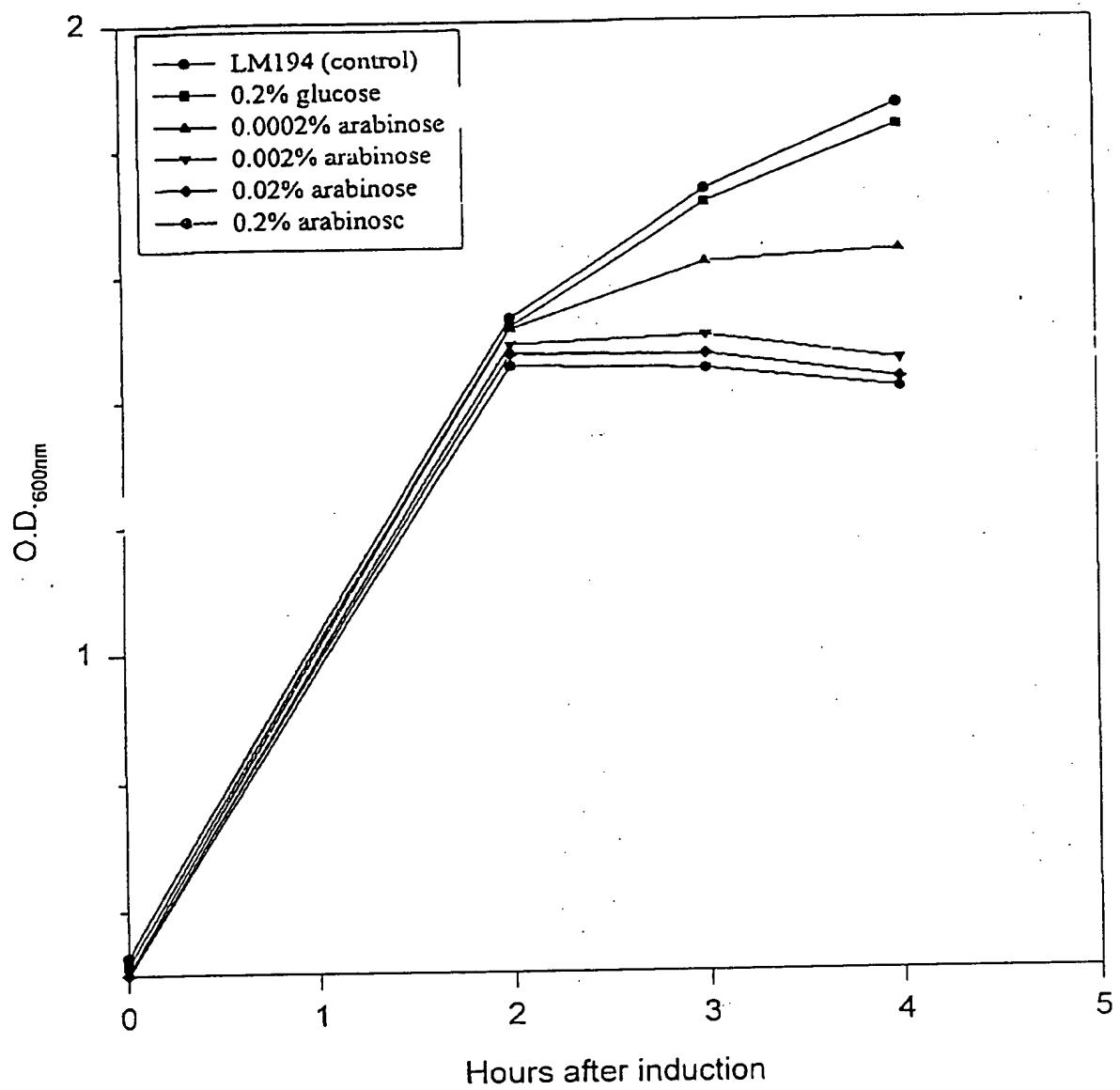


Fig. 16

### $\beta$ -lactamase in periplasmic space over time

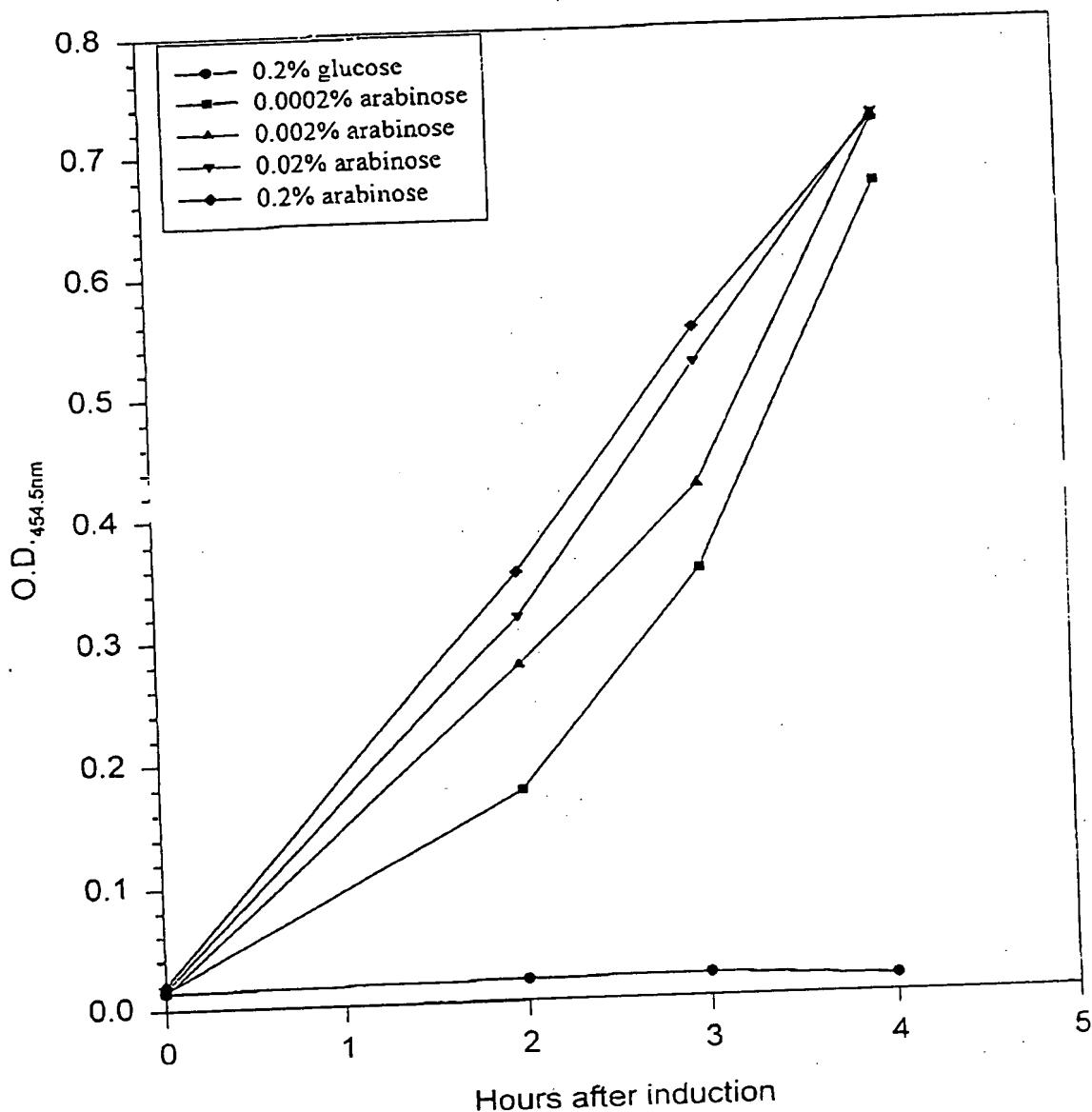


Fig. 17

### $\beta$ -lactamase in culture medium over time

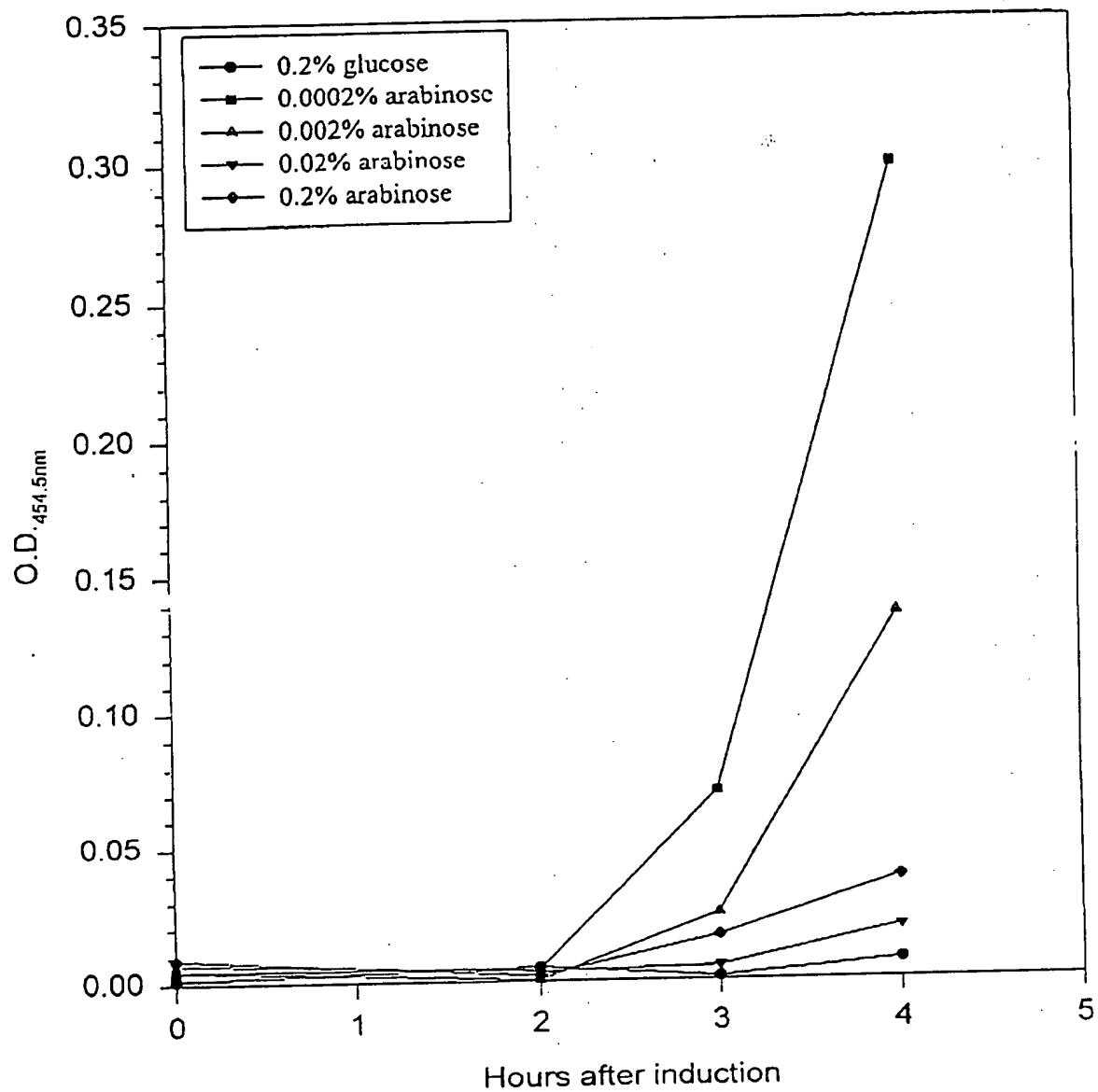


Fig. 18

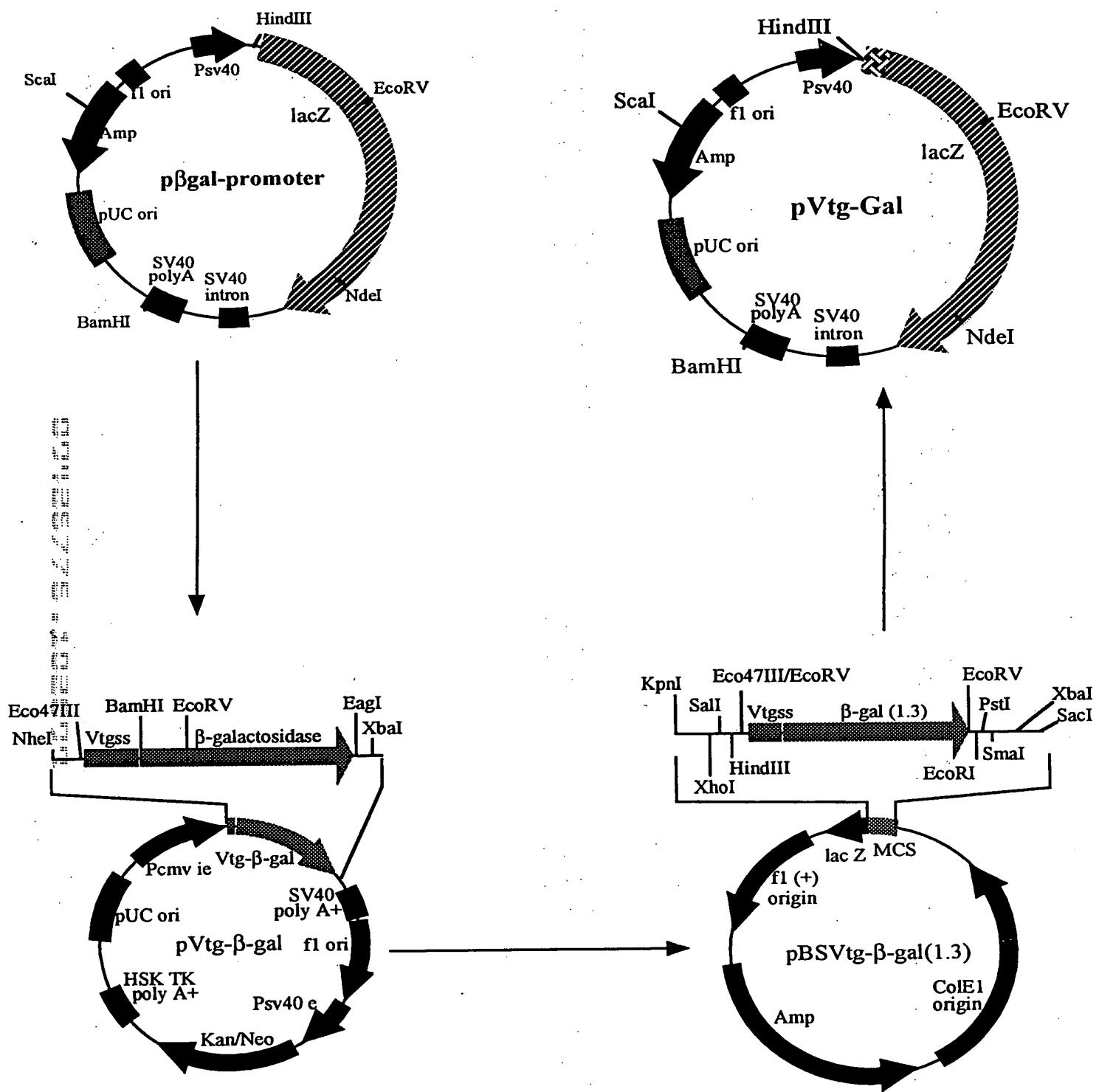


Fig. 19

Figure 21

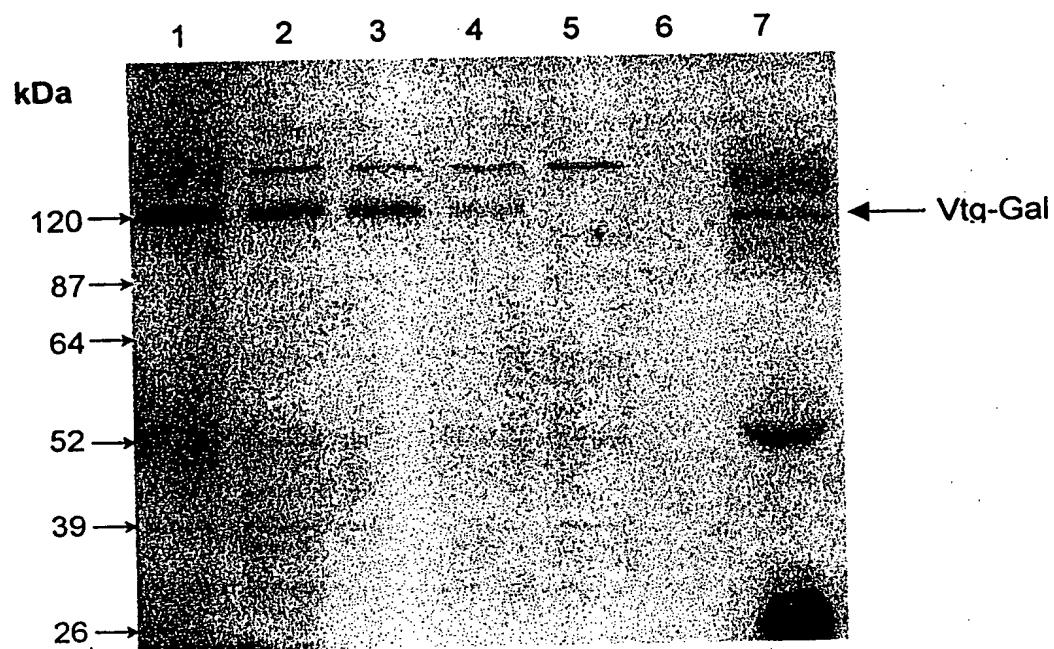


Fig. 20

## Rate of Secretion

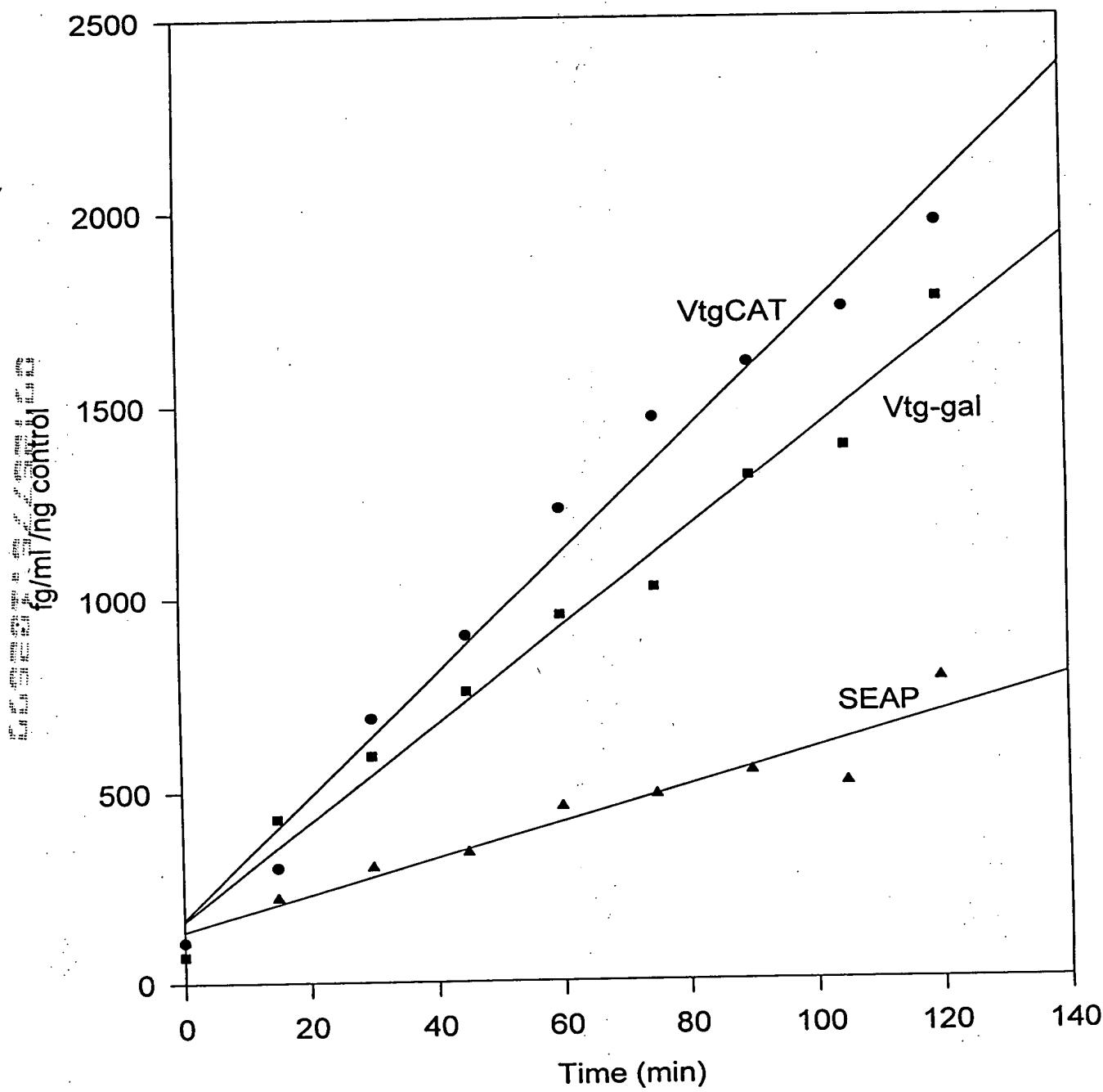


Fig. 21